CLAIMS

1. A method of producing double low restorer lines of Brassica napus for Ogura cytoplasmic male sterility (cms) presenting radish introgression carrying the Rfo restorer gene deleted of the radish Pgi-2 allele and recombined with the Pgi-2 gene from Brassica oleracea, and having a good agronomic value characterised by female fertility, a good transmission rate of Rfo and a high vegetative vigour, said method including the step of:
   a) crossing double low cms lines of spring Brassica napus comprising a deleted radish insertion with the double low line of spring Drakkar for forming heterozygous restored plants of Brassica napus,
   b) irradiating before meiosis the heterozygous restored plants obtained in step a) with gamma ray irradiation,
   c) crossing pollen from flowers obtained in step b) with the cms double low spring Wesroona line,
   d) testing the progeny for vigour, female fertility and transmission rate of the cms gene,
   e) selecting progeny lines.

2. A method according to claim 1, wherein the irradiation dose in step b) is 65 Gray during 6 mn.

3. A method according to claim 1 wherein the double low cms line of spring Brassica napus of step a) is R211.

4. A method according to claim 1 wherein the testing in step d) is performed with the combination of five markers selected from PGIol, PGIUNT, PGIint, BolJon and CP418.

5. Double low restorer lines of Brassica napus for Ogura cytoplasmic male sterility (cms) presenting a Rfo insertion deleted of the radish Pgi-2 allele and recombined with the Pgi-2 gene from Brassica oleracea, and having a

good agronomic value characterised by female fertility, a good transmission rate of Rfo and a high vegetative vigour.

6. Double low restorer lines of Brassica napus according to claim 5, wherein they present a unique combination of five markers selected from PGIol, PGIUNT, PGIint, BolJon and CP418.

7. Brassica napus hybrid plants and progeny thereof obtained through the steps of:

a) providing a restorer line produced according to claim 1 and bred to be homozygous,

b) using said restorer line in a hybrid production field as the pollinator,

c) using cms sterile plants in a hybrid production field as the hybrid seed producing plant, and

d) harvesting the hybrid seed from the male sterile plant.

8. The seeds of Brassica plant developed from the Brassica line obtained in claim 1.

9. The seeds of Brassica napus obtained in claim 7.

10. The seeds of Brassica napus obtained in claims 1 and 2 deposited in NCIMB Limited, 23 St Machar Drive, Aberdeen, Scotland, AB24 3RY, UK, on July 4 , 2003, under the reference number NCIMB41183.

11. Use of the combination of at least four markers PGIol, PGIint, BolJon and CP418, or any portion of them comprising at least one polymorphic site, for characterising recombined restorer lines of Brassica napus for Ogura cms presenting a Rfo insertion deleted of the radish Pgi-2 allele and recombined with the Pgi-2 gene from Brassica oleracea, and having a good agronomic value characterised by female fertility, a good transmission rate of Rfo and a high vegetative vigour.

12. Use according to claim 11 wherein the combination is of five markers PGIol, PGIUNT, PGIint, BolJon and CP418.

13. Use according to claim 12, wherein:

- The marker PGIol is amplified using the primers: PGIol U and PGIol L (PGIol U: 5'TCATTTGATTGTTGCGCCTG3'; PGIol L: 5'TGTACATCAGACCCGGTAGAAAA3')
- The marker PGIint is amplified using the primers: PGIint U and PGIint L (PGIint U: 5'CAGCACTAATCTTGCGGTATG3'; PGIint L: 5'CAATAACCCTAAAAGCACCTG3')
- The marker PGIUNT is amplified using the primers: PGIol U and PGIint L: (PGIol U: 5'TCATTTGATTGTTGCGCCTG3'; PGIint L: 5'CAATAACCCTAAAAGCACCTG3')
- The marker BolJon is amplified using the primers: BolJon U and BolJon L: (BolJon U: 5'GATCCGATTCTTCTCCTGTTG3'; BolJon L: 5'GCCTACTCCTCAAATCACTCT3')
- The marker CP418 is amplified using the primers: SG129 U and pCP418 L: (SG129 U: cf Giancola et al (5) pCP418 L : 5'AATTTCTCCATCACAAGGACC3')

14. PGIol marker whose sequence follows:

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TCATTTGATT GTTGCGCCTG TCGCCTTGTT GTGTTATGAT GAATGAACAG CAGTCATTTA  60
ACATGTGGTT AACTTAACAG GGCTCCGGCT GTTGCAAAAC ACATGGTTGC TGTCAGCACT 120
AATCTTGCGG TATGAATTTG TGATTAAATT TGTTTGTTTG TGACTCTTTC TTCATTGTTC 180
GTTTTCGTAC AATAAACCGA ATGTATAATC TTTTTACAAA CTGAATTTTC TACCGGGTCT 240
GATGTACA                                                          248
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15. PGIUNT marker whose sequence follows:

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TCATTTGATT GTTGCGCCTG TCGCCTTGTT GTGTTATGAT GAATGAACAG CAGTCATTTA  60
ACATGTGGTT AACTTAACAG GGCTCCGGCT GTTGCAAAAC ACATGGTTGC TGTCAGCACT 120
AATCTTGCGG TATGAATTTG TGATTAAATT TGTTTGTTTG TGACTCTTTC TTCATTGTTC 180
GTTTTCGTAC AATAAACCGA ATGTATAATC TTTTACAAAC TGAATTTTCT ACCGGGTCTG 240
ATGTACAATG CTAGTCTCCA TGTTCTTGGG GATCATGATT TATTTTCTAC ATGTATTCAG 300
ACAGTACAGA AGAAAGTGTT CAAAACTCTG GATGTTTTAA TTTACAGTTA GTGGAGAAGT 360
TCGGCATTGA TCCGAACAAT GCATTTGCAT TTTGGGACTG GGTTGGTGGA AGGTACAGTG 420
GTAAGTGCTT GTTTATTTGG TTGTATAAAT TTCTCGTCCA TTTCCGCTTG CTTAGTGTAT 480
AACTGAAATT CTTTTGCAGT TTGCAGTGCT GTTGGAGTCT TACCATTGTC TCTACAGTAT 540
GGCTTCTCTG TGGTTGAGAA GTACGGTACC TTCTACTTTA TCAGCCATCT CATAAAATGT 600
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CTTAGGCATA TTCTTTCTAT TTTATTTCCC TCTTAATGAT TTCTTCTTTT TTTTATTGCA 660
TTCCCGTTTT ATTTTCAAAA GTTGTTACTG TCTCTAAATC AAGAAGAAAC CTTCTTAGTA 720
GATCCAGCTG ATATTCAGCC TTTTTTAAAT TGGACTGCAG GTTTTTAAAG GGGAGCTTCA 780
AGCATTGATA AGCATTTCCA GTCCACACCG TTTGAGAAGA ATATACCCGT GAGTTGCATT 840
AGTTGTGTGA TTATACAGTT TTCTTGTCTT TTTGCTATGT CCATCAACAC TAGAGATTCG 900
TGAAGTTATT AGTGTAGTCA ACGCATAGGG AGAGGTGATT GGTGACTTTT GGACGATTTC 960
AGGTGCTTTA GGGTTATTG 979

16. PGIint marker whose sequence follows:

CAGCACTAAT CTTGCGGTAT GAATTTGTGA TTAAATTTGT TTGTTTGTGA CTCTTTCTTC 60
ATTGTTCGTT TTCGTACAAT AAACCGAATG TATAATCTTT TACAAACTGA ATTTTCTACC 120
GGGTCTGATG TACAATGCTA GTCTCCATGT TCTTGGGGAT CATGATTTAT TTTCTACATG 180
TATTCAGACA GTACAGAAGA AAGTGTTCAA AACTCTGGAT GTTTTAATTT ACAGTTAGTG 240
GAGAAGTTCG GCATTGATCC GAACAATGCA TTTGCATTTT GGGACTGGGT TGGTGGAAGG 300
TACAGTGGTA AGTGCTTGTT TATTTGGTTG TATAAATTTC TCGTCCATTT CCGCTTGCTT 360
AGTGTATAAC TGAAATTCTT TTGCAGTTTG CAGTGCTGTT GGAGTCTTAC CATTGTCTCT 420
ACAGTATGGC TTCTCTGTGG TTGAGAAGTA CGGTACCTTC TACTTTATCA GCCATCTCAT 480
AAAATGTCTT AGGCATATTC TTTCTATTTT ATTTCCCTCT TAATGATTTC TTCTTTTTTT 540
TATTGCATTC CCGTTTTATT TTCAAAAGTT GTTACTGTCT CTAAATCAAG AAGAAACCTT 600
CTTAGTAGAT CCAGCTGATA TTCAGCCTTT TTTAAATTGG ACTGCAGGTT TTTAAAGGGG 660
AGCTTCAAGC ATTGATAAGC ATTTCCAGTC CACACCGTTT GAGAAGAATA TACCCGTGAG 720
TTGCATTAGT TGTGTGATTA TACAGTTTTC TTGTCTTTTT GCTATGTCCA TCAACACTAG 780
AGATTCGTGA AGTTATTAGT GTAGTCAACG CATAGGGAGA GGTGATTGGT GACTTTTGGA 840
CGATTTCAGG TGCTTTAGGG TTATTG 866

17. BolJon marker whose sequence follows:

GATCCGATTC TTCTCCTGTT GAGATCAGCT CCAAACATCA AACAACTTGT ACACAAATAT 60
CTTTACTTGC TAAATGGAAC ATGACAAGAG ATAGAAAATC TTGCTCATAG TATTGTACAA 120
GGGATAACAG TGTAGAAAAC AAACCGTCTG TAAGATTTTC TCCCTGATCC TCTCACTTAA 180
CCAGTAGGCG TTTTTCACAT TGAAGCGCAT ATCTACTTTG GTATTCACTG AATAAAAAAA 240
GAAAGCTGGT AACATGTGAA GGATATACAA GCATTGATAC ACCAAGTAGT CACAAACTAC 300
ATTATAAAGG TCAGACCTTT GTTCACATTC TGGCCTCCAG GACCACCGCT TCTAGCAAAG 360
TTAAGCGTAA CATGGTCTGC ACGTATACAA ATGAAAATGT TTCTATCAAA ATCCTATAAA 420
ATAGAGCTCT ATAACATTGT CGATACATAG TTTCACTAAC TCTGCAAGTA CTAAACACAT 480
ATACAAACAA AACTATGCGA ACAGATCAAA ACTACTACAG AACACAGTTC TATGACACTG 540
TCGATAGTAA CATCCTCTGC AAGTACCAAA GAGATAGCAA ATGAAACTAT GTAAACAAAT 600
CAAAATTCTA AATTTCTCCA TCACAAGGAC CTACAGAATA GAGTTATCAT AACATTTTCT 660
GTAAATATTT CCATCAAAAT GACTAGAGAA CAGAGTTCTT ATAACATTAT CTGTAAATGT 720
TCCAACAAAA CCACTACATA GCAGAGTTCT TATAACATTG TCTGTAAATG TCCAATCAAA 780
ACCACTACAG AACAAAGCTC CTATAACATT GTTTATACAA AGTTTCACTA AATCTACAAA 840
CTTTCCCCGT AAATGAGCTT AATATCACCC AAAGATGTTT CAATCAGATA AAGAGTACGA 900
CATCGTTTTG AGATTAGAAC AAACTGAAAC TTACGTAGAG TGATTTGAGG AGTAGGC 957

18. CP418 marker whose sequence follows:

AATTTCTCCA TCACAAGGAC CTACAGAATA GAGTTATCAT AACATTTTCT GTAAATATTT 60
CCATCAAAAT GACTAGAGAA CAGAGTTCTT ATAACATTAT CTGTAAATGT TCCAACAAAA 120
CCACTACATA GCAGAGTTCT TATAACATTG TCTGTAAATG TCCAATCAAA ACCACTACAG 180
AACAAAGCTC CTATAACATT GTTTATACAA AGTTTCACTA AATCTACAAA CTTTCCCCGT 240
AAATGAGCTT AATATCACCC AAAGATGTTT CAATCAGATA AAGAGTAACG ACATCGTTTT 300
GAGATTAGAA CAAACTGAAA CTTACGTAGA GTGATTTGAG GAGTAGGCTC GTTGCCAGCA 360
GAGCTAGCTC TCTCCTCCGC CTCATGAAGC ATCTGTTGCA CCTGAGACAA CCGTGACGAA 420
ACTTTCCGAT CACCGCCACC AGAATTCGAC GCCGCGCATC GGAAGGATCC GAATCGGGAA 480
CTGAGTGAAC CCGAGCGATC CCGGGAGTGC GACGGAGCGA TGGGAAAAGA GAGTGGCACG 540
ATTTCGACGA AGAGTGGAAG AGGAGAGGGT GGTGGATAAA CTCGCGTATG ATCAAGTTCG 600
TCATCGTCCT GATTGCCGCC ATTTTTTTTG TCAGGGCGCT CTGTGGCTTA GAAGTTTCCG 660
atgtcaatga ac 672